Home Office

NON-TECHNICAL SUMMARY

Molecular mechanisms controlling immune cell killing

Project duration

5 years 0 months

Project purpose

- (a) Basic research

Key words

Immunology, Cancer, Therapy

Animal types	Life stages
Mice	Embryo and egg, Neonate, Juvenile, Adult, Pregnant adult

Retrospective assessment

> The Secretary of State has determined that a retrospective assessment of this licence is not required.

Objectives and benefits

Description of the projects objectives, for example the scientific unknowns or clinical or scientific needs it's addressing.

What's the aim of this project?

The aim of this project is to understand the molecular mechanisms controlling killer cells of the immune system.

Potential benefits likely to derive from the project, for example how science might be advanced or how humans, animals or the environment might benefit - these could be short-term benefits within the duration of the project or long-term benefits that accrue after the project has finished.

Why is it important to undertake this work?

Killer T cells are white blood cells of the immune system that defend our bodies against viral infections and cancers. There are 5 million killer T cells (also known as cytotoxic T lymphocytes) in every teaspoon of our blood. Recent advances in immunotherapies work by harnessing the ability of killer T cells to combat cancers. Understanding exactly how these killer cells work will provide vital information that will help to improve these transformative therapies.

What outputs do you think you will see at the end of this project?

We will identify the key pathways used by killer cells to deliver their lethal hit to cancer cells. We will publish our findings in peer-reviewed, open-access journals. Findings will be presented to the scientific community at conferences and to the wider lay audience via outreach activities including short videos available on the internet.

Who or what will benefit from these outputs, and how?

In the short term the scientific community will benefit from our discoveries that reveal how killer cells deliver their lethal hit. In the longer term many different cancer patients will benefit from discoveries that enable us to develop more efficient killer cells for treatment of many different cancers.

How will you look to maximise the outputs of this work?

We collaborate with clinicians, other scientists and pharmaceutical companies to maximise outputs from our research. We give talks to both specialist audiences as well as lay audiences including visits to schools and taking part in the Cambridge Science Festival. We have found that making short videos that explain our scientific discoveries and are hosted on the University of Cambridge's YouTube channel is very effective in communicating the outputs from our work to a broad audience. To date, our videos have accumulated >38 million views.

Species and numbers of animals expected to be used

- Mice: 3300

Predicted harms

Typical procedures done to animals, for example injections or surgical procedures, including duration of the experiment and number of procedures.

Explain why you are using these types of animals and your choice of life stages.

Adult mice enable us to generate the greatest number of mature killer cells per mouse, thereby using fewer mice. Adult mice are required to generate and preserve new genetically altered lines. Embryos and eggs are used to produce new lines of mice by fertilisation with sperm.

Typically, what will be done to an animal used in your project?

Typically, animals in this project will be bred and maintained for up to 6 months. They will be humanely killed to remove the spleen so that killer cells can be expanded in vitro. Occasionally, these mice may be used to generate new genetically altered lines, to cryopreserve these lines and to allow them to be housed in high health status animal units.

What are the expected impacts and/or adverse effects for the animals during your project?

Genetically altered animals being bred are not expected to exhibit any adverse effects. Animals are monitored every day and will be humanely killed immediately if they exhibit any adverse effects.

Animals undergoing surgical procedures may experience transient post-operative discomfort and can be given pain relief. Animals undergoing non-surgical embryo transfer will experience no more than mild transient discomfort and no lasting harm.

Expected severity categories and the proportion of animals in each category, per species.

What are the expected severities and the proportion of animals in each category (per animal type)?

Mice: Mild 94%; Moderate 6%

What will happen to animals used in this project?

- Kept alive at a licensed establishment for non-regulated purposes or possible reuse
- Killed
- Used in other projects

Replacement

State what non-animal alternatives are available in this field, which alternatives you have considered and why they cannot be used for this purpose.

Why do you need to use animals to achieve the aim of your project?

The parallels between mouse and human are well understood and mice are known to provide excellent models of human disease. This allows us to study mutations that cannot be studied in man and generate results that are directly relevant to humans. Cells isolated from mice only survive in culture for 4 weeks, requiring continued use of mice. We continue to advance the use of improved in vitro cell culture, genetic manipulation and human cell lines as viable alternatives.

Which non-animal alternatives did you consider for use in this project?

Mouse and human cell lines have been tested as alternatives for this project. Artificially produced T cells produced in vitro were also considered. We will continue to test new non-animal alternatives.

Why were they not suitable?

Cell lines that grow in culture long term contain viruses and no longer accurately reflect the function of primary killer T cells that function in vivo and artifically produced killer T cells do not faithfully acquire all of the properties of killer T cells. Whole animals are needed to provide bona fide killer cells as they acquire their unique characteristics as they develop inside the animal.

Reduction

Explain how the numbers of animals for this project were determined. Describe steps that have been taken to reduce animal numbers, and principles used to design studies. Describe practices that are used throughout the project to minimise numbers consistent with scientific objectives, if any. These may include e.g. pilot studies, computer modelling, sharing of tissue and reuse.

How have you estimated the numbers of animals you will use?

In the last year we have used just under 600 mice; thus we estimate we will use 3000 mice over the next 5 years. These numbers are calculated as the minimum number of mice in a colony to maintain breeding and allow for 4 mice per week to be used to isolate killer T cells from each colony.

What steps did you take during the experimental design phase to reduce the number of animals being used in this project?

After we isolate killer T cells from the spleens of mice we increase the number of killer T cells from each mouse by growing them in a dish, in liquid that contains nutrients that promote cell growth. This means that we can reduce the number of mice needed. As we are working with the cells isolated from mice, randomisation and blinding are not required. If we do need to ensure statistical experiments we consult with specialist statisticians.

What measures, apart from good experimental design, will you use to optimise the number of animals you plan to use in your project?

We have optimised our methods to maximise the number of killer cells that we can generate in the laboratory by growing them in a dish with appropriate nutrients. This means that we need fewer mice to generate killer cells for our studies. Where possible we modify the genetic material in the cells we have grown in the laboratory so that we do not need to breed additional genetically modified mice.

Refinement

Give examples of the specific measures (e.g., increased monitoring, post-operative care, pain management, training of animals) to be taken, in relation to the procedures, to minimise welfare costs (harms) to the animals. Describe the mechanisms in place to take up emerging refinement techniques during the lifetime of the project.

Which animal models and methods will you use during this project? Explain why these models and methods cause the least pain, suffering, distress, or lasting harm to the animals.

We will use genetically modified mouse strains that do not develop any adverse effects. Surgical proceedures are carried out by experienced staff in the facility to ensure the least pain and suffering to the animals.

Why can’t you use animals that are less sentient?

In this project we only use animals to produce the cells required for experiments that we conduct in dishes in the laboratory. No animals are used in experiments, just their cells and tissues. Animals such as flies, fish and frogs have different ways of fighting off infection.

How will you refine the procedures you're using to minimise the welfare costs (harms) for the animals?

Where possible, non-surgical procedures are used during implantation (Protocol 3). Any animals undergoing surgical procedures will be carefully monitored using welfare scoring sheets. Animals wo;; be maintained in a high helath status unit so that they are free from diseases. Trained staff look after the animals and check on them every day. The cages have environmental enrichment for the animals and nesting materials. Animals are handled via the cupped hand method, or using their tunnels. When, very occasionally, surgery is required injections are known not to have caused more than mild and transient adverse effects, and surgery is carried out aseptically. Animals are killed using a humane method.

What published best practice guidance will you follow to ensure experiments are conducted in the most refined way?

The department provides continually updated guidance on experimental refinements. Use of the website from the NC3Rs (https://www.nc3rs.org.uk) in the 3Rs resource library for in vivo techniques as well as husbandry, and LASA (Laboratory Animal Science Association) will also be made.

How will you stay informed about advances in the 3Rs, and implement these advances effectively, during the project?

We continually review methods for replacement, reduction and refinement provided by the scientific literature and our support team within the facility, attending workshops to stay up to date. Named information officers provide a valuable source of 3Rs information. We also make use ot the NCR3 website.